does detect interaction between the two proteins using the yeast two-hybrid system (Leber et al., 2001), raising a question about the binding of Cvt19 solely to the presequence of prAPI.

Supporting the premise that Cvt19 is the prAPI receptor is the finding that Cvt19 is degraded in the vacuole with similar kinetics as the processing of prAPI, and that its turnover is dependent on other components of the Cvt pathway and the vacuolar proteinase encoded by the *PEP4* gene (Scott et al., 2001). Cvt19 is also needed for the specific and rapid vacuolar delivery of API by macroautophagy under starvation conditions but not for the slower, nonspecific delivery of API to vacuoles (Leber et al., 2001).

One surprise is that unlike most receptors that act in multiple rounds of binding and release, Cvt19 is unorthodox in that it self-destructs in the vacuole while delivering API there. This seems like a costly price for specificity, but perhaps the inefficiency is mitigated by the fact that multiple dodecamers of prAPI (and also Ams1 oligomers) may be delivered to the vacuole by each Cvt19 molecule. Another unexpected result is that although there is genetic evidence for a role of Cvt19 in Ams1 delivery to the vacuole, no interaction was detected between Cvt19 and Ams1 (Scott et al., 2001). It is possible that the low abundance of Ams1 makes such an interaction difficult to detect, but an alternative possibility compatible with the genetic data and the absence of a presequence in Ams1 is that the Cvt complex, including Ams1, may interact with Cvt19 solely via prAPI. This point also raises the question of what other cargoes might need Cvt19 and the Cvt pathway for vacuolar delivery.

Since the Cvt pathway is constitutive in vegetative cells, do Cvt vesicles participate in a futile cycle of formation in the cytosol and degradation in the vacuole in the absence of cargo? Some light is shed on this problem by the finding that the vacuolar turnover of Cvt19 is API dependent (Scott et al., 2001), but the broader question remains in the absence of complete knowledge of all Cvt cargoes.

We are also left with some other unanswered questions. What protein and/or lipid confers upon Cvt19 its binding specificity for the punctate, vesicular structures near the vacuole? What is the nature and origin of these structures? Is the proximity of these structures to the vacuole reflective of their organelle of origin or of their final site of consumption at the vacuole? Somewhat surprisingly, even in the absence of Cvt19 multimeric prAPI associates with membranous structures, albeit less strongly than in wild-type cells, raising the possibility that some other protein and/or lipid functions with Cvt19 as either upstream- or co-receptors for prAPI (Scott et al., 2001). The rapid progress in this field promises that answers to these queries and perhaps a uniform gene nomenclature system will be forthcoming.

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Fas-Acting Memory

Genetic and behavioral analysis points to a role for fasciclin II in controlling odor memory and alcohol sensitivity in *Drosophila*.

Regulation of cell-adhesion molecules can bring about alterations in synaptic structure that are plausibly associated with long-term changes in memory. Surprisingly, some of these adhesion molecules are also implicated in immediate learning.

It is likely that forming new memories involves changes in the efficacy of individual synapses in specific neuronal circuits. Evidence from many systems implicates second-messenger systems in these synaptic changes. Current models envisage that short-term memories are stored as labile electrophysiological changes at synapses, whereas long-term memories are encoded as structural alterations at the same synapses. Many studies have indicated a role for cell-adhesion molecules in the long-term, morphological type of synaptic change (Martin and Kandel, 1996). However, the true picture may not be that simple. Studies with the *Drosophila* memory mutant *volado*, whose gene encodes an α-integrin subunit, demonstrated that cell-adhesion molecules can also be critical for short-term memories (Grotewiel et al., 1998). A recent paper in *Cell* extends this finding by implicating yet another *Drosophila* cell-adhesion molecule, fasciclin II (FasII), in short-term memory formation (Cheng et al., 2001).

Many *Drosophila* learning genes are highly expressed in the adult fly mushroom bodies (MBs)—brain structures that are necessary for olfactory learning (Zars, 2000). The Davis group has previously screened *Drosophila* P element enhancer-trap lines to find genes that are expressed at high levels in the MBs (see Cheng et

al., 2001 for a list). Several of these genes are required for olfactory learning. The latest gene announced from this screen is *fasII* (Cheng et al., 2001). Mutant *fasII* flies have an olfactory learning deficit.

fasII comes from an interesting gene family. Its relatives include mammalian neural cell adhesion molecule (N-CAM) and *Aplysia* ApCAM (Martin and Kandel, 1996). N-CAM and ApCAM have been implicated in long-term synaptic plasticity. All genes of this family encode both transmembrane and glycophosphatidylinositol (GPI)-anchored variants.

The developmental role of *Drosophila fasII* has been studied for several years. The name of the gene comes from the aberrant axonal fasciculation observed in *fasII* mutant embryos (Lin et al., 1994). Later, Schuster et al. (1996), studying the larval neuromuscular junction (NMJ), showed that *fasII* downregulation is necessary for activity-dependent synaptic growth and for the increase in synaptic sprouting and efficacy generated by elevated cAMP (see also Martin and Kandel, 1996). Informed by these earlier studies, the gross and fine anatomy of the adult MBs were examined. Overall structure of the MBs appears normal in the weakest $fasII^{rd2}$ mutant, as does the cellular organization of the MB lobes in electron-microscopic sections. This apparent CNS normality is in contrast to the larval NMJ, which is demonstrably altered in *fasII* mutants. Perhaps differences in CNS synapses are simply harder to detect in weak mutants.

The original *fasII* mutant flies that Cheng et al. identified were weak and reproduced poorly. They generated two new alleles that were less sluggish. All these mutant alleles are predicted to affect the expression of all three FasII forms, two transmembrane, and one GPI anchored. They tested the healthier *fasII* mutants for a learning defect. Mutant *fasII* flies have a modest reduction in immediate olfactory memory. Importantly, the mutant flies can detect both the odor and the electric shock stimuli that are required for olfactory learning. So *fasII* mutants are impaired in learning. How do we know that the gene affects learning acutely rather than chronically by affecting neural development? This was addressed by temporally controlling expression of a *fasII* transgene using the heat shock promoter.

They introduced a heat shock promoter-driven *fasII* transgene, encoding a transmembrane FasII isoform, into *fasII* mutant flies. The transgene fully restored wild-type learning to *fasII* mutant flies. However, this rescue was independent of heat shock. Apparently the transgene promoter was leaky—*fasII* was expressed even at 25°C. To circumvent this problem, the authors manipulated the system—the *fasII* transgene could be switched off if the flies were placed at 18°C, but it could be induced by incubating the flies at 25°C. Using this, Cheng et al. demonstrated that rescue of *fasII* mutant memory is reversible. If the flies were kept overnight at 18°C prior to testing, their memory performance was as low as *fasII* mutant flies. Induction of the *fasII* transgene in these flies by 1–2 hr incubation at 25°C restored wild-type memory. Finally, if the flies were returned to 18°C, the olfactory memory returned to mutant levels. These data strongly imply that transmembrane FasII function is acutely required for adult olfactory learning. The authors suggest that the rapid shift and reversibility makes it unlikely that synapses have developed improperly in *fasII* mutants. Raising the *fasII* mutants without *fasII* transgene induction (i.e., at 18°C) and then inducing the gene only in adults (by shifting them to 25°C) before testing would better answer this question. Nevertheless, the learning deficit of *fasII* mutant flies is the same as that of *fasII* mutant flies raised with the *fasII* transgene on, but then switched off in adulthood.

The involvement of FasII in memory formation versus memory retrieval was addressed by testing the memory of *fasII* mutant flies that have the *fasII* transgene induced before or after learning. Whereas *fasII* expression prior to learning restores wild-type memory, providing *fasII* expression after learning does not. This finding implies that *fasII* is required for memory formation but does not discount that it might also be required for recall.

Some genes that affect associative learning are also involved in the response of flies to ethanol exposure (Moore et al., 1998 and see Waddell and Quinn, 2001). Cheng et al. add *fasII* flies to the list of ethanol sensitive learning mutants. In addition, they show that the transgene conditions that rescue olfactory memory do not restore normal ethanol sensitivity. In fact, *fasII* overexpression can worsen the sensitivity. This might suggest that the cellular response to learning and ethanol exposure are different, as has been proposed previously (DeZazzo et al., 1999). However, as the authors note, it is possible that a normal ethanol response also requires the GPI-anchored form of FasII.

A critical question for cell adhesion molecules in short-term memory remains: do they function by triggering cell-signaling cascades or are they just sticking and unsticking? Cheng et al. suggest that perhaps short-term memory is mediated by a signaling function whereas long-term memory involves a cell adhesion role. The answer to this may come "fas"t.

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